

## Chemical composition and origin source of soil organic matter under four vegetation in Doñana National Park

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### Abstract:

This study deals with the chemical characterization of soil organic matter (SOM) under different ground covers from a Mediterranean climate (Andalusia, South Spain), for it using techniques of analytical pyrolysis, with this techniques, we could find that organic matter consists mainly of seven chemical families, as they are, alkanes / alkenes, fatty acids, aromatic, lignin, steranes, sugars and peptides. Also we had been used different approaches for the isotopic signature study of stable light elements in bulk (low-complexity) samples. Light element isotope ratios ( $\delta^{15}\text{N}$ ,  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$  and  $\delta\text{D}$ ) were measured in the whole soil samples (EA/TC-IRMS) and  $\delta^{13}\text{C}$  and  $\delta\text{D}$  values were also estimated in a number of specific compounds -previously identified by Py-GC/MS released after pyrolysis (Py-GC-(FID)-C\TC-IRMS).

### Introduction

Among the most suitable techniques for the direct study of complex organic matrices such is soil organic matter (SOM) is analytical pyrolysis. The technique consists of a thermolytic degradation of macromolecules into small fragments that may be separated and identified by gas chromatography–mass spectrometry (Py-GC/MS). Pyrolysis of SOM generates a wide range of products that can be related to their origin (e.g., methoxyphenols from lignin, anhydrosugars and furan derivatives from polysaccharides, and N-containing molecules from proteins; González-Vila et al., 2001; Leinweber & Schulten, 1995). In addition, recently the technique has been effectively hyphenated with other detection devices like isotope ratio mass spectrometry (IRMS) that will provide additional information relevant for the monitoring of biogeochemical processes including tracing the origin and dynamics of SOM pools.

In this study we use different approaches for the isotopic signature study of stable light elements in bulk (low-complexity) arenosol samples collected under different vegetation covers from a Mediterranean climate (Andalusia, South Spain). Light element isotope ratios ( $\delta^{15}\text{N}$ ,  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$  and  $\delta\text{D}$ ) were measured in the whole soil samples (EA/TC-IRMS) and  $\delta^{13}\text{C}$  and  $\delta\text{D}$  values were also estimated in a number of specific compounds -previously identified by Py-GC/MS- released after pyrolysis (Py-GC-(FID)-C\TC-IRMS).

### Experimental

The Soil samples were collected in a circular area (radius 5m) under frequent vegetation covers found in sandy soils from the Doñana National Park (SW Spain): cork oak (*Quercus suber*, QS), eagle fern (*Pteridium aquilinum*, PA), pine (*Pinus pinea*, PP) and rockrose (*Halimium halimifolium*, HH). Dry soil

samples were sieved to fine earth (< 2 mm) to discard coarse elements and litter.

Pyrolysis-gas chromatography–mass spectrometry (Py-GC/MS) was performed for SOM characterization using a double-shot pyrolyzer (Frontier Laboratories, model 2020i) attached to a GC/MS system Agilent 6890N. Compound assignment was achieved via single-ion monitoring for various homologous series, via low-resolution mass spectrometry, and comparison with published and stored (NIST and Wiley libraries) data.

Bulk isotopic signature of light elements ( $\delta^{15}\text{N}$ ,  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$  and  $\delta\text{D}$ ) was analyzed using a Flash 2000 HT (N, C, S, H and O) elemental analyzer coupled to a Delta V Advantage IRMS (Thermo Scientific) (EA/TC-IRMS).

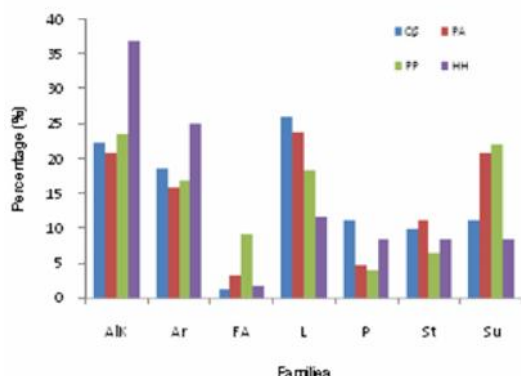
The direct study of specific compounds isotopic signature of light elements ( $\delta^{13}\text{C}$  and  $\delta\text{D}$ ) was done by coupling a pyrolysis unit (double-shot pyrolyzer “Frontier Laboratories, model EGA/Py-3030D”) – to a gas chromatograph fitted with a flame ionization detector (GC/FID) and coupled to the Delta V Advantage IRMS (Thermo Scientific GC-Isolink System) (Py-GC-(FID)-C\TC-IRMS).

Isotopic ratios are reported as parts per thousand (‰) deviations from appropriate standards recognized by the international atomic energy agency (IAEA) (Valkiers et al., 2007).

### Results and Discussion

#### Pyrolysis characterization:

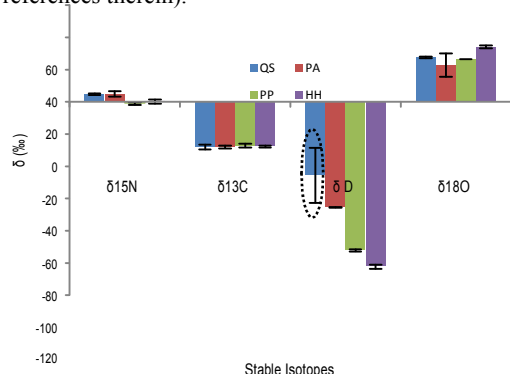
The organic matter in each soil sample presented rich pyrolysates with a large number of chemical compounds. These could be best classified in seven groups according to its chemical nature or probable biogenic origin (Fig 1)



**Figure 1.** Distribution (relative abundance) of main chemical families identified in SOM pyrolysates (ALK: Alkane/Alkene; Ar: Aromatic; FA: Fatty Acid; L: Lignin; P: Peptides; St: Steranes and Su: Sugars) under the different studied vegetation types (QS: *Quercus suber*; PA: *Pteridium aquilinum*; PP: *Pinus pinea* and HH: *Halimium halimifolium*).

#### EA/TC-IRMS:

Bulk soil isotopic signatures of light elements in each sample are shown in Fig 2. The  $\delta^{13}\text{C}$  signature is clearly in the range of C3 plant (-26 to -30 ‰) (O'Leary 1981) and the different plant canopies (tree, shrubs or ferns) caused only slight variations in  $\delta^{13}\text{C}$  (STD=0.42). Nitrogen isotope signature ( $\delta^{15}\text{N}$ ) is also in line with that commonly found in plant or land organisms as described in Létolle (1980). Cross plots of  $\delta^{15}\text{N}$  vs.  $\delta^{18}\text{O}$  may provide information about nitrate ( $\text{NO}_3^-$ ) sources and N cycling (Kendall, 1998), in our case, it was compatible with a predominant nitrate source from atmospheric deposition ( $\delta^{15}\text{N}$  range: -5 to 5 ‰;  $\delta^{18}\text{O}$  range: 20 to 70 ‰). No conclusive results could be obtained from the  $\delta\text{D}$  isotopic signature probably due to overlapping of the  $\delta\text{D}$  signals from the organic and the mineral fractions. For a more accurate  $\delta\text{D}$  analysis additional steps allowing their separation would be necessary (Ruppenthal et al., 2013, and references therein).

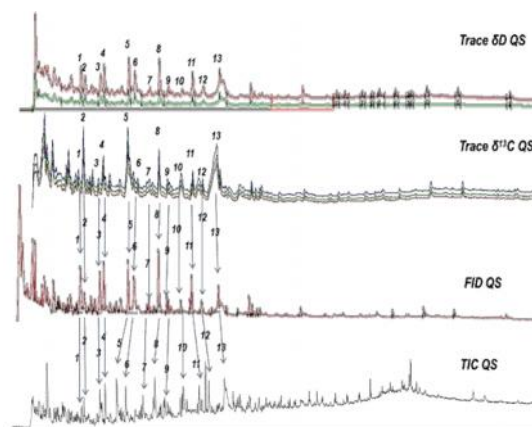


**Figure 2.** Bulk isotopic signature of light elements under the different studied vegetation types (*Quercus suber* QS, *Pteridium aquilinum* PA, *Pinus pinea* PP and *Halimium halimifolium* HH).

#### Py-GC-(FID)-C\TC-IRMS:

An example of the compound specific IRMS analysis of selected peaks released directly from

pyrolysis is depicted in Fig. 3 and Table 1. These results are actually in the process of analysis and discussion. The results presented during the meeting will be discussed in relation to possible different sources contributing to SOM as well as to specific micro-climatic conditions that may be affecting SOM isotopic signature at a local scale.



**Figure 3.** Example of compound specific ( $\delta^{13}\text{C}$  and  $\delta\text{D}$ ) analysis (Py-GC-(FID)-C\TC-IRMS) of soil under *Quercus suber* (QS). Numbers on traces corresponds to compounds of known structure as determined by conventional analytical pyrolysis (TIC QS: Py-GC/MS) and listed in Table 1.

**Table 1.** Isotopic signature of light elements in selected peaks released directly from pyrolysis of soil under *Quercus suber* (QS) (Py-GC-C\TC-IRMS).

N°	Name	Family	$\delta\text{D}$ (‰)	$\delta^{13}\text{C}$ (‰)
1	Trimetil beneno	AROMATIC	-65.93	-27.58
2	1-Undecene	AIKENE	-68.81	-27.51
3	Phenol	AROMATIC	-73.51	-27.52
4	Guaiacol	LIGNIN	-75.91	-26.56
5	Phenol, 4-methyl-	AROMATIC	-65.06	-26.66
6	Phenol, 2-methoxy-4-methyl-	LIGNIN	-47.84	-26.80
7	Phenol, 4-ethyl-2-methoxy-	LIGNIN	-64.41	-27.47
8	2-Methoxy-4-vinylphenol	LIGNIN	-62.41	-27.99
9	Syringol	LIGNIN	-78.90	-27.47
10	2H-Pyran-2,4(3H)-dione, 3-acetyl-6-methyl-	SUGARS	-63.61	-27.24
11	Cyclopentanone, 2-(1-methylpropyl)-	SUGARS	-77.81	-28.42
12	Vinylsyringol	LIGNIN	-55.44	-28.52
13	Levoglucozan	SUGARS	-43.34	-26.87
Bulk isotopic signature of QS samples (Mean±STD)			-45.68±17.07	-27.98±1.48

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